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JOINT HIGHWAY RESEARCH PROJECT

FHWA/IN/JHRP-82/9

EFFECTS OF FERTILIZATION PRACTICES DURING NURSERY PRODUCTION ON MYCORRHIZAL DEVELOPMENT BY LANDSCAPE PLANTS USED FOR HIGHWAY REVEGETATION

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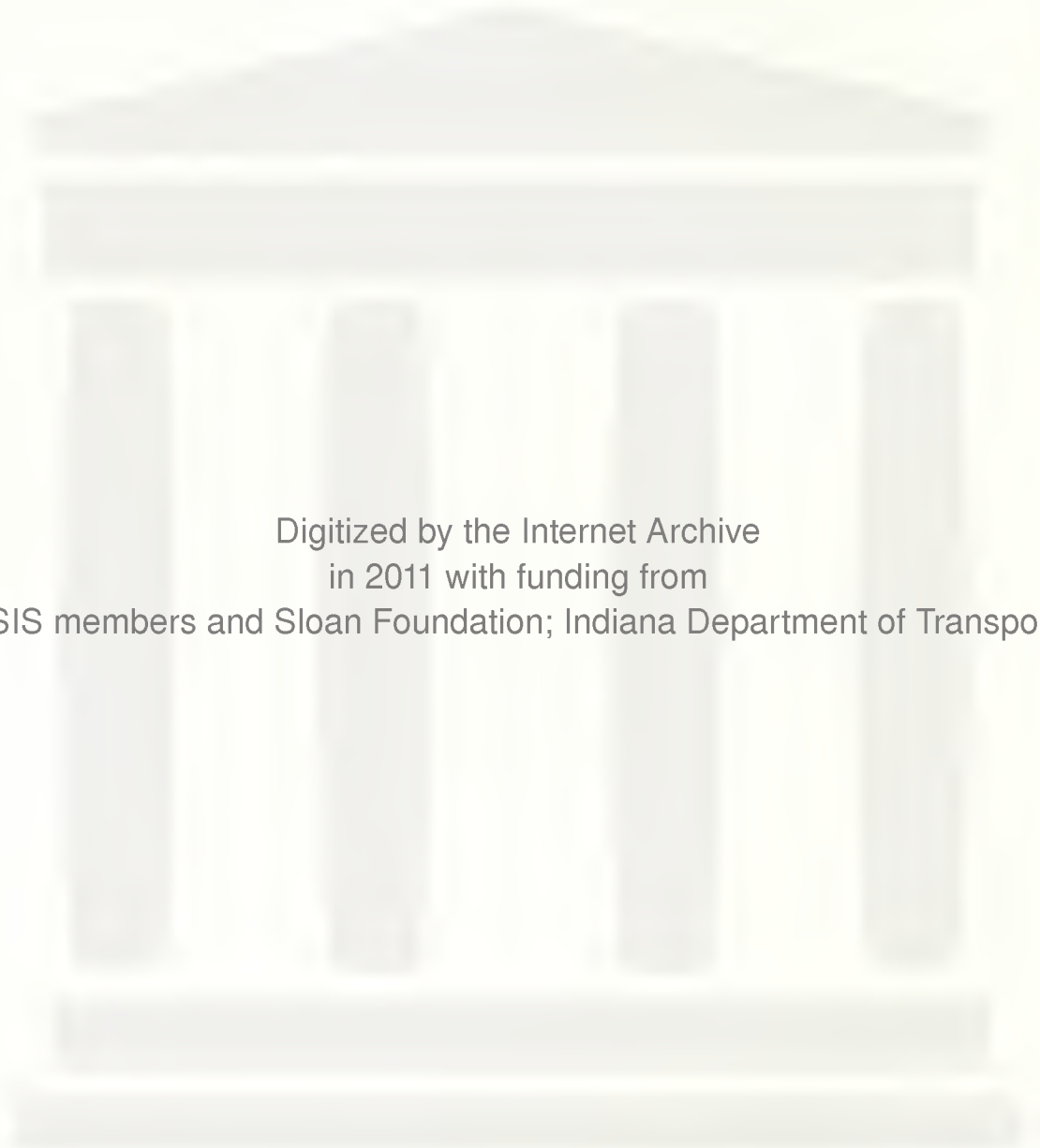

Interim Report

EFFECTS OF FERTILIZATION PRACTICES DURING NURSERY PRODUCTION ON MYCORRHIZAL DEVELOPMENT BY LANDSCAPE PLANTS USED FOR HIGHWAY REVEGETATION

TO: H. L. Michael, Director
Joint Highway Research Project

April 6, 1982

Project: C-36-48H

FROM: David F. Hamilton

File: 9-5-8

Attached is an Interim Report on the HPR Part II Study titled "Techniques to Increase Survival of New Highway Plantings". This Interim Report is the second one on the portion of the Study concerned with use of mycorrhizal fungi. The Report is titled "Effects of Fertilization Practices During Nursery Production on Mycorrhizal Development by Landscape Plants Used for Highway Revegetation".

The Report is submitted for review and acceptance as partial fulfillment of the objectives of the Study. A few photographs in a few copies will be in color as this better illustrates the matter illustrated. Most copies as published, however, will not have color prints as they make the publication cost high.

The Final Report on this portion of the Study is in preparation and will be available very soon.

Respectfully submitted,

David F. Hamilton/dfh

David F. Hamilton

DFH:ms

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INTERIM REFPORT

EFFECTS OF FERTILIZATION PRACTICES DURING NURSERY PRODUCTION ON MYCORRHIZAL DEVELOPMENT BY LANDSCAPE PLANTS USED FOR HIGHWAY REVEGETATION

by

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16. Abstract

Effects of endomycorrhizal inoculation on growth of selected landscape plants fertilized at different nutrient levels were determined. Liriodendron tulipifera, Forsythia x intermedia, Acer platanoides, and Lolium perenne were grown in 3.28 liter containers under greenhouse conditions. Thirty-nine plants of each species were inoculated either with Glomus fasciculatus or Glomus mosseae. For each plant species, inoculated and noninoculated plants were treated with 0, 2, or 4 g/l Nitrogen of 19N-6P-12K controlled release fertilizer.

The optimum fertility level for most plant species studied was 2 g/l N. Mycorrhizal development increased growth of L. tulipifera infected with G. fasciculatus at both 2 and 4 g/l N. When no supplemental fertilizer was applied, there was no growth increase of mycorrhizal plants. Growth of A. platanoides and L. perenne was not improved by mycorrhizal association with G. fasciculatus, and growth of F. x intermedia was not improved by association with G. mosseae. However, according to other researchers different fungal symbionts may increase growth of these plants.

Successful inoculation during production would yield plant material for use on highway sites which already has mycorrhizal development. Results indicate that if the correct combination of plant species and mycorrhizal fungi are selected, mycorrhizal development can be achieved during production despite conditions of high fertility.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

HIGHLIGHT SUMMARY

In nature and under cultivation, many horticultural plants are capable of forming symbiotic mycorrhizal associations. Mycorrhizae benefit plant growth mainly through increased nutrient and water uptake. When soil microbes are not present, as in sterile media or soils of highly disturbed landscape sites, plant growth may be slowed.

Since a commercial nurseryman must earn a profit from producing landscape plants, efficiency is essential. Production costs, particularly for fertilizers, are rising rapidly as inflation increases. As crop production intensifies, nurserymen use high applications of fertilizers to enhance plant growth. Container production of nursery crops relies greatly on the use of media without soil or the normal microflora needed for root growth. Also, soils at highly disturbed sites, such as reclaimed strip-mine and highway slopes, have very low levels of beneficial soil microbes and are difficult to revegetate.

The introduction of mycorrhizal fungi to the rhizosphere of plants grown in containers may make it possible to use soil nutrients more efficiently and reduce production costs. Mycorrhizal roots also may increase growth and survival of these plants once they are transplanted to the landscape, particularly on harsh sites.

Although many plants are known to have mycorrhizal symbionts which increase height and plant weight, effects of mycorrhizal development on plant growth over a range of fertility levels are unknown. The objective of this study was to determine the effects of mycorrhizal inoculation and soil fertility on the growth of selected landscape plants used for revegetating Indiana's highways.

The right combination of plant species and mycorrhizal fungi are essential if mycorrhizal infection is to be beneficial. The development of mycorrhizal plants during production could eliminate the need for inoculation at planting.

The optimum fertility level for tulip poplar was at lower levels of fertilization although mycorrhizal development increased growth at both low and high rates of fertilization. Mycorrhizal development did not promote growth of tulip poplar without supplemental fertilizer.

INTRODUCTION

Soils present in the planting zones of highways generally provide disturbed and harsh environments for successful long-term establishment of vegetation. Unfavorable aspects of these highway locations often include extremes of environment especially high temperatures, low or excess moisture, very limited nutrient supplies (low fertility) and a lack of beneficial microorganisms. Often these planting soils are subsoils or exposed hardpans.

A previous interim report showed that many highway soils had no mycorrhizal spores present, while successfully revegetated highway soils had large populations of mycorrhizal fungi. Mycorrhizae promote growth of plants in several ways including enhanced nutrient uptake, thus resulting increased growth and survival.

The previous interm report also showed that plant species may not be compatible with all species of mycorrhizal fungi. While growth of some plants was shown to be enhanced by mycorrhizal symbiosis, no responses to optimum fertilizer-mycorrhizal fungi interactions was determined for other species.

Effects of Mycorrhizae on Soil Nutrient Use

Inoculation with endomycorrhizal fungi result in more efficient use of soil nutrients by many landscape plants, and increasing plant size (Hayman, 1978). Mycorrhizal plants have an advantage under conditions of low fertility, due to the absorptive surface added to the root system by the fungal hyphae and perhaps to more efficient absorption (Harley, 1971). Mycorrhizal development is even reduced under conditions of high fertility, with some plant species (Menge et al., 1978; Maronek et al., 1980; Mosse, 1973; Johnson et al., 1980; Powell, 1980). There are very few reports of increased growth of mycorrhizal plants at high fertility levels (Meyer, 1974; Wright, 1971).

Mycorrhizal promotion of plant growth is very species and nutrient specific. Inoculation of Liquidambar styraciflua with a mixture of Glomus mosseae and Glomus etunicatus enhanced plant growth from 140 to 1120 kg/ha of 10N-10P-10K (Schultz et al., 1979; Kormanik et al., 1977). Glomus fasciculatus increased growth of several citrus cultivars over a wide range of fertility (Menge et al., 1978). Glomus fasciculatus also promoted growth of avocado plants (Persea americana Mill.) over a wide range of fertility, except when zinc was withheld and phosphorus was present in large amounts (Menge et al., 1980).

Height of Magnolia grandiflora was promoted by inoculation with G. fasciculatus at 1.1 kg/m^3 and 4.5 kg/m^3 (Maronek et al., 1980). Fresh weights of roots and shoots of Podocarpus macrophyllus Thunb., and Rhododendron simsii Planch. inoculated with a mixture of G. mosseae and G. fasciculatus were increased when fertilized with 250 to 1250 mg/l N and K with 50 to 250 mg/l Mg (Johnson et al., 1980).

Uptake of several nutrients is also reported to be improved by endomycorrhizal formation (Gerdemann, 1965; Mosse, 1973), but these reports are inconsistent. Nitrogen uptake has been enhanced by mycorrhizae in soybeans (Glycine max L.) (Ross and Harper, 1970; Ross, 1971), and ericaceous plants (Read and Stribley, 1973; Stribley and Read, 1980). Stribley and Read (1980) report that mycorrhizae utilize nitrogen more effectively by using simple organic compounds not used by nonmycorrhizal plants. Mycorrhizal roots may also have increased nitrate-reducing capacity (Ho and Trappe, 1975).

Uptake of sulfur by mycorrhizal onion (Rhodes and Gerdemann, 1978), red clover (Trifolium pratense L.), and maize (Zea mays L.) was also higher than by nonmycorrhizal plants. In addition, uptake of zinc was greater by mycorrhizal, than by nonmycorrhizal Malus speciosa Mill. (Benson and Covey, 1976). There are also some reports of increased uptake of K, Ca, Mg, Fe, Cu, Mn, Na, and B

by mycorrhizal plants (Gerdemann 1965).

Reports of increased phosphorus uptake by mycorrhizal plants have been more numerous than for any other nutrient. In fact, enhancement of plant growth by mycorrhizae occurs particularly under conditions of low soil phosphorus (Hayman, 1978; Mosse, 1973; Stribley et al., 1980). Increases in P concentration, rather than total P content in the plant, have been reported for tomato (Lycopersicon esculentum L.) (Cress et al., 1979), subterranean clover (Trifolium subterrareum L.), cassava (Manihot esculenta Mill.), soybean, and maize (Stribley et al., 1980). However, Schultz et al. found decreases in concentrations of N, P, K, and Mg in mycorrhizal L. styraciflua.

As mycorrhizal plants increase in size, their total P content rises proportionately, but the P concentration of shoots or roots may remain the same as that of nonmycorrhizal plants. Species with greater total P content include mycorrhizal tulip poplar (Gray and Gerdemann, 1967), perennial ryegrass (Powell, 1977), onion (Allium cepa L.) (Gray and Gerdemann, 1967; Hattingh et al., 1973), and alfalfa (Medicago sativa L.) (Barea et al., 1980). Other reports have not been consistent and mycorrhizal plants either have higher, equal, or lower P concentrations than nonmycorrhizal plants. This most likely depends on the availability of the nutrients and rate of plant growth.

One explanation for increased nutrient uptake is that the mycorrhizal hyphae act as fine roots or root hairs (Baylis, 1972), and simply serve as an additional, well distributed surface for absorbing nutrients (Mosse, 1973; Hattingh et al., 1973; Sanders and Tinker, 1973). Other authors suggest the possibility that not only are mycorrhizal root systems more extensive, but they are also more effective for nutrient uptake. Cress et al. (1979) reported that increased P uptake may be due to a greater affinity of mycorrhizal absorbing sites for $H_2PO_4^-$. It is a widespread view that mycorrhizae can solubilize

unavailable phosphate, but Mosse (1973) reported that mycorrhizal plants used the same P sources as nonmycorrhizal plants. Furthermore, some nonmycorrhizal plants may have a minimum threshold concentration of soil P, below which they cannot absorb phosphate. According to this theory, mycorrhizal plants have a lower threshold or no threshold at all.

The varied growth response of mycorrhizal plants to phosphorus may be controlled by two opposing factors (Harley, 1969; Stribley et al., 1980). There is a stimulating effect due to enhanced P uptake, and a detrimental effect due to the fungal drain on host photosynthate. Therefore, if the plant is adequately supplied with P, the plant can actually suffer a yield loss in the presence of mycorrhizae (Crush, 1975; Stribley et al., 1980). According to Gerdemann (1965), if the nutrient is limiting plant growth, it may occur in higher concentrations in the mycorrhizal plant. If a mycorrhizal plant does not reach its full growth potential relative to P availability, due to a lack of other nutrients or a fixed carbon loss, the plant will have a greater percent P and a lower dry weight.

It is essential that installation and maintenance costs for plant materials be kept as low as possible along highways and other highly-disturbed sites. Installation costs, maintenance costs (including fertilization) and replacement costs have historically been expensive because the harsh soil and site conditions make successful establishment difficult without a high maintenance input.

The introduction of mycorrhizal fungi to the rhizosphere of plants during production or at transplanting may make it possible to use soil nutrients more efficiently and reduce maintenance and replacement costs through the increased growth and survival of the plants, especially on harsh sites. Plants selected are frequently planted along Indiana highways. The objectives of this study were: a) to determine the effects of mycorrhizal inoculation on growth of

selected plants under a range of fertility levels; b) to determine if Glomus mosseae is a mycorrhizal symbiont of tulip poplar and to compare this fungal species to G. fasciculatus; c) to determine if G. fasciculatus and G. mosseae develop mycorrhizae on tulip poplar and are beneficial under highly fertile production conditions.

MATERIALS AND METHODS

Experiment 1.

Seeds of tulip poplar (Liriodendron tulipifera L.) and Norway maple Acer platanoides L.) were planted in perlite and sand (1:1,v/v) and grown for 4 weeks in a growth chamber at 26 $\pm$ 2^{o}C under a 13 hour photoperiod. Terminal cuttings of forsythia (Forsythia x intermedia Zab.) (20 cm long with 8 leaves) were rooted in perlite and vermiculite (1:1, v/v) under intermittent mist (12 sec./10 min.) in a greenhouse with approximately 25% shade. Indolebutyric acid (IBA) was used at 0.1% (w/v) to enhance rooting. Rooted cuttings of forsythia, seedlings of tulip poplar (4 cm tall), and Norway maple (4 to 8 cm tall) were transplanted in 3.28 liter pots (one gallon trade designation) containing steam pasteurized medium of perlite, sphagnum peat moss, and soil (2;2:1, v/v/v). Seeds of perennial ryegrass (Lolium perenne L.) were seeded directly into 3.28 liter pots at a rate of 12.33 g/m^2, and covered with 0.5 cm of pasteurized medium. Before addition of fertilizer, the medium for all plants contained 9 gm/l N, 1 mg/l P, and 4 mg/l K. The pH was 5.5 - 6.5.

Half the plants of each species were inoculated either with Glomus fasciculatus (Thaxter) Gerd. and Trappe or Glomus mosseae Nicol. & Gerd., at a rate of 44,000 spores/m^2 of soil surface area (Table 1). The inoculum contained tomato roots (Lycopersicon esculentum L.), fungal spores (Figure I), hyphae, and the growing medium (perlite:sphagnum peat moss:soil (2:2:1, v/v/v).

The inoculum was inserted into three vertical cores around the plant to a depth of 10 cm in the pot.

Inoculated and noninoculated plants were grown with fertilizer additions of 0, 2, or 4 g/l N of medium, supplied by controlled release `Osmocote' fertilizer[1] (19N-6P-12K) in a 3 to 4 month release period. As N was increased, P and K were increased proportionally. Fertilizer was incorporated to a depth of 5 cm. Plants were grown in a greenhouse (24 $\pm$ 3°C under a 16 hour photoperiod) and watered as needed with tap water.

After three months the plants were harvested and the roots and shoots were separated. Measurements included shoot length, dry weights of roots and shoots, total root length (Tennant, 1975), and the percentages of N, P, and K in roots and shoots. The percentage of N was determined by Nesslerization, P by the ammonium-phospho-molybdate method with 1,2,4-amino napthol sulphonic acid as the reducing agent (Jackson, 1958), and K by flame spectrophotometry with a model 9200 Unicam flame spectrophotometer. Nutrient concentration of roots and total root length were determined for all species except perennial ryegrass. Shoot length was measured for all species except forsythia and perennial ryegrass. The amount of mycorrhizal infection was determined by visual estimation using root staining and microscopy (Gray and Gerdemann, 1967; Phillips and Hayman, 1970; Giovanetti and Mosse, 1979). The treatments were arranged in a randomized complete block design with 13 replicates for height measurements and 6 replicates for all other measurements. Data were analyzed by analysis of variance, with the Newman-Keuls test of significance used to separate means.

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[1]Sierra Chemical Company, Milpitas, California.

Experiment 2.

Seeds of tulip tree were planted in a medium of perlite:vermiculite (1:1, v/v) and grown in a growth chamber ($75^{o}F \pm 2^{o}F$ day/$72^{o}F \pm 1^{o}F$ night, with a 16 hour photoperiod) for 5 weeks. Thirty seedlings, approximately 4 cm tall, were transplanted into 0.725 liter pots (one quart trade designation) containing steam pasteurized soil (perlite:peat moss:soil; 2:2:1, v/v/v). The unfertilized medium contained 120 ppm NO_3^-, 53 ppm P, 130 ppm K, 12 ppm N and had a pH of 6.2.

Ten plants were inoculated with G. fasciculatus (Thaxter) Gerdemann & Trappe, at a rate of 44,000 spores/m^2 of soil surface area, and ten with G. mosseae at the same rate. The inoculum contained soil, roots, fungal spores, and hyphae from the previous host. The inoculum was inserted in three vertical cores in the pot to a depth of 6 cm. The experiment was conducted in a greenhouse ($80^{o}F \pm 10^{o}F$, with a 14 hour photoperiod), and watered with nutrient water (200 ppm N, 200 ppm P, and pH 6.8) as needed. Height was measured at the beginning and end of the experiment. The experiment was in a completely randomized design.

RESULTS

Experiment 1.

Liriodendron tulipifera

Inoculation of L. tulipifera with viable G. fasciculatus resulted in mycorrhizal development (Table 2). Roots of inoculated plants grown at 2 or 4 g/l N had high mycorrhizal development (more than 50% of cortical cells infected with hyphae) (Figure 2). Inoculated plants grown without supplemental fertilizer (0 g/l N) had very little mycorrhizal development (less than 5% of cortical cells infected with hyphae). Noninoculated plants had no mycorrhizal infection.

Inoculation with G. fasciculatus increased plant growth of fertilized plants (Figure 3). Without supplemental fertility, there was no significant increase in the heights of plants with or without mycorrhizal inoculation. At 2 or 4 g/l N, inoculated plants were taller than noninoculated plants, but there was no significant difference between the two fertility levels.

Mycorrhizal inoculation also increased the dry weights of shoots from fertilized plants. Without fertilization, there was no difference between the dry weights of shoots from inoculated and noninoculated plants. At 2 and 4 g/l N, shoots of inoculated plants were significantly heavier than shoots of non-inoculated plants. Dry weights of shoots from inoculated plants fertilized with 4 g/l N were also greater than dry weights of shoots from inoculated plants fertilized with 2 g/l N. Dry weights of noninoculated plants did not differ significantly regardless of fertility level.

Inoculation also increased the dry weights of roots from fertilized plants. There was no difference between the dry weights of roots from inoculated and noninoculated plants grown without fertilizer. At 2 and 4 g/l N, dry weights of roots from inoculated plants were heavier than those from noninoculated plants. However, there was no difference between the dry weights of roots from plants grown at the two fertility levels.

Mycorrhizae also increased root lengths of fertilized plants. Inoculated plants grown at 2 or 4 g/l N had longer roots than those from other treatments, but were not significantly different from each other. Root length of inoculated plants grown without fertilizer and noninoculated plants grown at all nutrient levels did not differ.

Finally, there were differences in the nutrient concentrations of inoculated and noninoculated plants at the three fertility levels (Table 3). Inoculation did not affect foliar N concentrations, although foliar N

concentration was increased by additions of fertilizer. Inoculation slightly increased the percentage of N in roots of plants grown at 2 or 4 g/l N. Nitrogen in roots also was less in both inoculated and noninoculated plants grown without fertilizer.

Foliar P concentration was the same for all treatments except noninoculated plants grown without supplemental fertilizer, which had less phosphorus than the other treatments. Therefore, mycorrhizal development increased the P concentration in shoots of inoculated plants grown without supplemental fertilizer. Phosphorus concentrations were greatest in noninoculated roots from plants grown at 2 g/l N.

Inoculation increased foliar K concentration of unfertilized plants. However, the highest foliar K concentration was in noninoculated plants grown at 4 g/l N. Mycorrhizal development did not affect the concentration of K in roots. The percentage of K in roots was significantly less in plants grown without fertilizer.

Acer platanoides:

Inoculation of A. platanoides with G. fasciculatus resulted in minimal mycorrhizal development (Table 2). The least mycorrhizal development on inoculated plants occurred when no fertilizer was added. High fertility also suppressed mycorrhizal development, since infection was greatest at 2 g/l N. However, even at 2 g/l N mycorrhizal development was not great. Noninoculated plants had no mycorrhizal development.

Although inoculation resulted in some mycorrhizal development, it did not enhance plant growth at any fertility level studied (Table 4). Addition of fertilizer did increase growth. Height, dry weights of shoots and roots, and root length were the same for all treatments except plants grown without supplemental fertilizer, which had less growth than other treatments.

Increasing fertility from 2 to 4 g/l N did not further increase growth.

While inoculation did not promote overall growth, there were some effects on plant nutrient concentrations (Table 5). Inoculation and increases in fertilizer application from 2 to 4 g/l N did not significantly affect the N concentration in roots and shoots of inoculated plants, there was a trend of increased N concentration in shoots and a trend of decreased N concentration in roots of inoculated plants.

Foliar P concentration of plants grown at 2 or 4 g/l N decreased following inoculation. However, inoculation increased the P concentration in roots of plants at these fertility levels.

Mycorrhizal inoculation also did not affect K concentration in roots or shoots of Norway maple. Potassium concentration was least when no fertilizer was applied (0 g/l N), but raising the fertility level from 2 to 4 g/l N did not affect percentage of potassium in roots or shoots.

Forsythia x intermedia:

Mycorrhizal inoculation of forsythia with G. mosseae did not result in infection, except for a small amount with inoculated plants grown without fertilizer (Table 2). Plants grown at 4 g/l N were heaviest, with unfertilized plants weighing the least (Table 6). Inoculation with G. mosseae did not significantly increase shoot weight, although inoculated plants tended to be heavier.

Dry weights of roots were not significantly influenced by inoculation, but were increased by fertilization. Fertilization with 2 g/l N increased root dry weight, but additional fertilizer application did not further increase growth.

Inoculation did not influence foliar N and P concentrations of forsythia, but did slightly decrease foliar K concentration (Table 7). Fertilization increased foliar N concentration, but increasing the fertilizer rate from 2 to

4 g/l N did not further increase N concentration.

Inoculation increased the N concentration in roots of fertilized plants (2 and 4 g/l N), but fertility had no effect on noninoculated plants. Inoculation also increased P concentration of roots at 0 or 2 g/l N. Phosphorus concentration was greatest in inoculated plants grown at 2 g/l N. Potassium concentration of roots was increased in fertilized plants, but was not further increased by additional fertilizer. Inoculation decreased K concentration in roots.

Root length was not significantly influenced by fertility or inoculation, although inoculation tended to increase root length of fertilized plants. Plants grown at 2 g/l N tended to have the greatest total root length, while plants grown at 0 g/l N tended to have the shortest total root length.

Lolium prenne:

Mycorrhizal development of L. perenne with G. fasciculatus was not extensive (Table 2). There was slight infection (less than 5% of cortical cells infected) on roots of all inoculated plants. Mycorrhizal infection was greatest at 2 g/l N and was less at higher or lower nutrient levels.

Dry weight of shoots was not influenced by inoculation (Table 8). Fertilization increased shoot growth, but raising fertilizer applications from 2 to 4 g/l N did not further improve growth. Foliar N was also unaffected by inoculation, although it was influenced by fertility. Nitrogen concentration was highest in plants grown at 2 g/l N. Foliar P concentration was increased by inoculation at 2 g/l N, and was lowest in unfertilized plants. Potassium concentration was increased by fertilization, but not affected by inoculation. Increasing fertility from 2 to 4 g/l N did not enhance foliar K concentration.

DISCUSSION

Experiment 1:

With the species tested, mycorrhizal inoculation promoted plant growth in tulip poplar infected with G. fasciculatus. Inoculation of tulip poplar was effective for increasing plant growth only under fertilized conditions. Plants grew taller with longer and heavier roots at both 2 or 4 g/l N, but shoots were heaviest at 4 g/l N. Without supplement fertilization at the growing medium, mycorrhizal inoculation was not beneficial to the host plant.

The data indicated that inoculated plants at 4 g/l N had greater shoot:root ratios than those of plants grown at 2 g/l N. This is indicated by the significant increase in dry weights of shoots of inoculated plants at 4 g/l N relative to those at 2 g/l N, but no similar increase for dry weights of roots. The altered shoot:root ratio may be caused by the increased nitrogen supply at the 4 g/l N level. An excessively high shoot:root ratio could prohibit establishment and survival of the plants in the landscape.

Mycorrhizae have a potential role in the container production of tulip poplar because of the increases in growth following inoculation. Establishment of an inoculation program can enable more efficient use of nutrients by plants.

However, for other plant species studied inoculation had no beneficial effect on plant growth at the fertility levels tested, even though slight mycorrhizal development was observed. Plants with roots which are better adapted for nutrient uptake may be less dependent on mycorrhizal formation, and therefore may have greater specificity for a particular fungal symbiont before increased growth occurs. Perhaps such plant-fungal associations would result in growth increases at fertility levels more marginal for plant growth than those studied (less than 2 g/l N but higher than 0 g/l N).

For all plant species, foliar and root nutrient concentrations were

influenced by inoculation, indicating definite interactions even when plant growth was not influenced. If growth is limited by any factor, it is feasible that mycorrhizae may still influence uptake of some nutrients, resulting in higher concentrations of those nutrients in the plant. However, if mycorrhizal development acts to reduce the effect of the limiting factor even slightly, then the concentration of some nutrients may be reduced as the nutrients are spread over a larger volume of plant tissue.

The optimum nutrient level appears to be 2 g/l N for the plants studied regardless of interactions with mycorrhizal fungi, with the exception of inoculated tulip poplar, which had heavier shoots when fertilized with 4 g/l N. Addition of fertilizer greater than 2 g/l N would not result in any growth increases for some plant species.

In plants which did not receive a growth stimulation from mycorrhizal formation, there was no detrimental effect from mycorrhizal inoculation. This suggests that when several plant species are being inoculated, an inoculum source could contain numerous species of mycorrhizal fungi to accommodate all plants present. Plants forming mycorrhizal roots without growth increases showed no inhibition of growth following inoculation under extremes of fertility conditions. However, in such cases where the plant and fungal species are not highly compatible, other environmental conditions, such as extreme temperature, may result in inhibition of growth of inoculated plants. Inoculum intended for various plant species should have at least one highly compatible fungal species for each plant species present, to insure increased growth.

In summary, it appears that minimum nutrient requirements must be met before inoculation with mycorrhizal fungi will result in increased growth. Inoculation of tulip poplar was beneficial to plant growth even under fertile conditions (4 g/l N of 19N-6P-12K), but the optimum fertilizer rate for most

species studied was 2 g/l N. Establishment of an inoculation program can enable more efficient use of nutrients by plants. For growth increases to occur, the plant and fungal symbionts must be compatible, and nutrients must be available. Knowledge of the economic feasibility of maintaining a pure inoculum source will help determine the commercial success of an inoculation program.

Experiment 2:

Both G. fasciculatus and G. mosseae were successful mycorrhizal symbionts of tulip poplar under highly fertile production conditions. Tulip poplars inoculated with G. fasciculatus (36.10 cm tall) and those inoculated with G. mosseae (28.19 cm tall) were significantly taller than noninoculated control plants (15.37 cm tall). Plants inoculated with G. fasciculatus tended to be taller than plants inoculated with G. mosseae, indicating a slight competative edge by G. fasciculatus.

These results indicate that if the correct combination of plant species and mycorrhizal fungi are selected, mycorrhizal development can be achieved during production despite high fertility rate. Use of these mycorrhizal plants could eliminate the need for mycorrhizal inoculation at highway sites during establishment, since mycorrhizal root systems have already been developed.

CONCLUSIONS

1. Minimum nutrient requirements must be met before inoculation with mycorrhizal fungi will result in increased growth. In other words, increases in growth of plants attributable to mycorrhizal development are greatest with some supplemental nutrient source.

2. Mycorrhizal development can occur under conditions of very low to very high fertility during production but plant responses may be limited (i.e. increased

growth). Inoculation during production is possible even with high rates of supplemental fertilizer. Such successful mycorrhizal inoculation during production will yield plants preadapted to transplanting and will be beneficial especially on difficult-to-revegetate sites.

3. For growth increases to occur, the plant and fungal symbionts must be compatible. It is possible to get limited mycorrhizal infection with no positive growth responses. This emphasizes further that growth responses due to mycorrhizal development may occur with only very specific fungi, even though infection can occur with several species of fungi.

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Table 1. Combinations of plant and fungal species used, and subsequent compatibility.

Plant Species	Mycorrhizal Fungi	Growth Increase
Acer platanoides	*Glomus fasciculatus*	None
Forsythia x intermedia	*Glomus mosseae*	None
Liriodendron tulipifera	*Glomus fasciculatus*	Positive
Lolium perenne	*Glomus fasciculatus*	None

Table 2. Percent of cortical cells infected with mycorrhizae in roots of inoculated and noninoculated L. tulipifera, A. platanoides, F. x intermedia, and L. perenne grown at three fertility levels.

Plant	Fertility (g/l N)	Inoculated	Noninoculated
L. tulipifera	0	0-2	0
	2	60-80	0
	4	60-80	0
A. platanoides	0	0-2	0
	2	0-2	0
	4	0-2	0
F. x intermedia	0	0-2	0
	2	0-2	0
	4	0-2	0
L. perenne	0	0-2	0
	2	0-2	0
	4	0-2	0

Table 3. Effects of fertility and inoculation with *Glomus fasciculatus* on nutrient concnetration (% N, P, and K) in roots and shoots of *Liriodendron tulipifera*.[z]

Nutrient (%)	Fertility Level (g N/l)	Shoots		Roots	
		Inoculated	Noninoculated	Inoculated	Noninoculated
N	0	1.069b	0.706v	0.418s	0.323d
	2	2.037a	2.584a	1.467c	1.183c
	4	2.329a	3.108a	2.025a	1.867ab
P	0	0.205a	0.062b	0.193b	0.198b
	2	0.157a	0.138a	0.413b	0.978a
	4	0.161a	0.228a	0.378b	0.201b
K	0	1.075c	0.578c	1.833b	1.870b
	2	1.438b	1.662b	2.973a	2.970a
	4	1.112bc	2.275a	2.063a	3.150a

[z] Separation of means by the Newman-Keuls test of significance, 5% level. Mean of 6 values. Differing letters represent significance in rows and columns for each nutrient in roots and shoots.

Table 4. Effects of fertility and inoculation with *Glomus fasciculatus* on growth of *Acer platanoides*.[z]

Plant	Fertility	Inoculated	Noninoculated
Height Increase (cm)[y]	0	0.64b	0.34b
	2	55.35a	43.41a
	4	49.32a	49.13a
Dry Weight of Shoots (g)[x]	0	0.343b	0.517b
	2	1.382a	1.270a
	4	1.890a	1.471a
Dry Weight of Roots (g)[x]	0	0.311b	0.309b
	2	7.283a	7.117a
	4	7.8967a	6.817a
Root length (cm)[x]	0	1297.0b	1290.6b
	2	5848.2a	8229.7a
	4	8724.9a	8227.7a

[z] Separation of means by the Newman-Keuls test of significance, 5% level. Differing letters represent significance in rows and columns for each plant response.

[y] Mean of 13 values.

[x] Mean of 6 values.

Table 5. Effects of fertility and inoculation with Glomus fasciculatus on nutrient concentration (% N, P, and K) in roots and shoots of Acer platanoides.[z]

Nutrient (%)	Fertility Level (g N/l)	Shoots		Roots	
		Inoculated	Noninoculated	Inoculated	Noninoculated
N	0	0.49b	0.81b	0.37c	0.44c
	2	2.43a	2.41a	1.64b	1.77ab
	4	2.72a	2.60a	1.69b	2.03a
P	0	0.09b	0.14b	0.11c	0.14bc
	2	0.18b	0.29a	0.24a	0.17b
	4	0.21b	0.34a	0.25a	0.18b
K	0	0.65b	0.94b	0.94b	1.30b
	2	1.75a	1.64a	1.93a	2.06a
	4	1.52a	1.65a	2.00a	1.95a

[z] Separation of means by the Newman-Keuls test of significance, 5% level. Mean of 6 values. Differing letters represent significance in rows and columns for each nutrient in roots and shoots.

Table 6. Effects of fertility and inoculation with *Glomus mosseae* on growth of *Forsythia* x *intermedia*.[z]

Plant Response	Fertility (g/l N)	Inoculated	Noninoculated
Dry Weight of Shoots (g)	0	0.750c	0.941c
	2	10.250b	9.123b
	4	14.328a	12.913a
Dry Weight of Roots (g)	0	1.099b	0.688b
	2	2.975a	3.140a
	4	3.874a	3.241a
Root Length (cm)	0	418.2a	614.2a
	2	4088.4a	3465.2a
	4	2425.5a	1579.3a

[z] Separation of means by the Newman-keuls test of significance, 5% level. Mean of 6 values. Letters represent significance in rows and columns for each plant response.

Table 7. Effects of fertility and inoculation with Glomus mosseae on nutrient concentration (% N, P, and K) in roots and shoots of Forsythia x intermedia.[z]

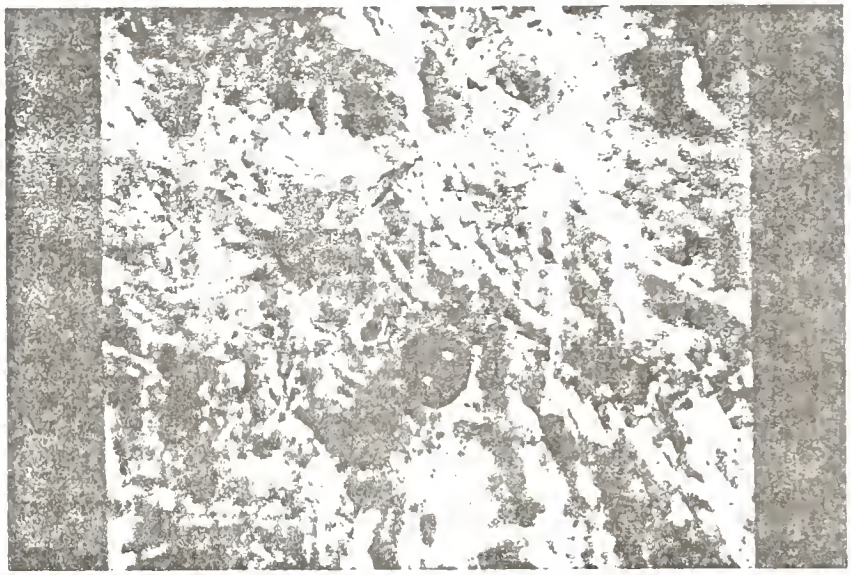
Nutrient (%)	Fertility Level (g N/l)	Shoots		Roots	
		Inoculated	Noninoculated	Inoculated	Noninoculated
N	0	0.458b	0.727b	0.858b	0.613b
	2	3.354a	3.342a	1.729ac	0.577b
	4	3.317a	3.185a	1.953a	0.577b
P	0	0.088b	0.087b	0.078c	0.032d
	2	0.250a	0.240a	0.203a	0.135b
	4	0.218a	0.243a	0.165b	0.146b
K	0	0.745s	0.792d	1.351c	1.388c
	2	2.184bc	2.318ab	2.563b	3.198a
	4	2.048cc	2.473a	2.563b	3.273a

[z] Separation of means by the Newman-Keuls test of significance, 5% level. mean of 6 values. Differing letters represent significance in rows and columns for each nutrient in roots and shoots.

Table 8. Effects of fertility and inoculation with *Glomus fasciculatus* on shoot dry weight (g) and nutrient concentration (% N, P, and K) of *Lolium perenne*.[z]

Plant Response	Fertility (g/l N)	Inoculated	Noninoculated
Shoot Dry Weight (g)	0	0.032b	0.035b
	2	0.977a	3.082a
	4	2.754a	3.012a
Foliar N (%)	0	1.353c	1.218c
	2	3.367a	3.533a
	4	3.112b	3.000b
Foliar P (%)	0	0.365c	0.344c
	2	0.575a	0.452b
	4	0.528a	0.547a
Foliar K (%)	0	1.953b	1.991b
	2	3.388a	3.532a
	4	3.185a	3.255a

[z] Separation of means by the Newman-Keuls test of significance, 5% level. Mean of 6 values. Differing letters represent significance in rows and columns for each plant response.

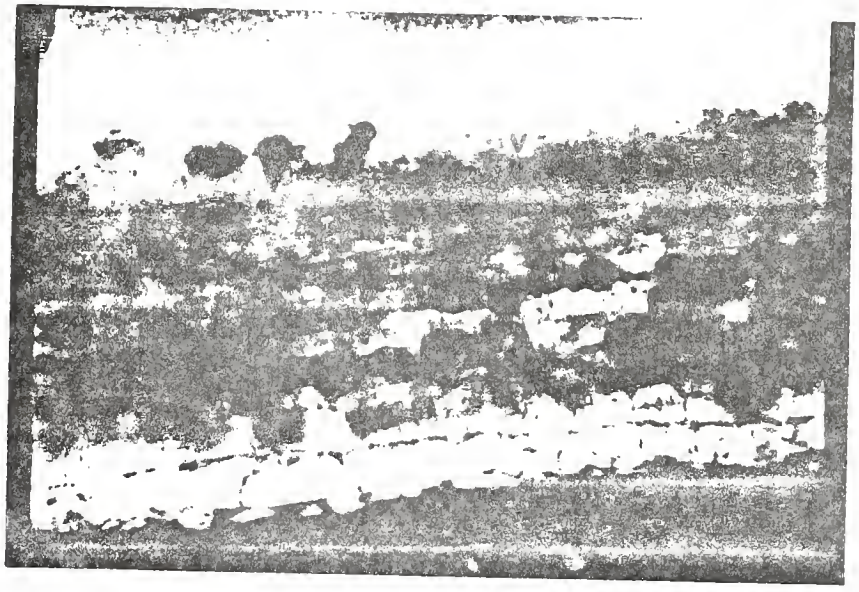


a. Glomus mosseae spore

b. Germinating Glomus fasciculatus spore

Figure 1. Glomus spores (72X)

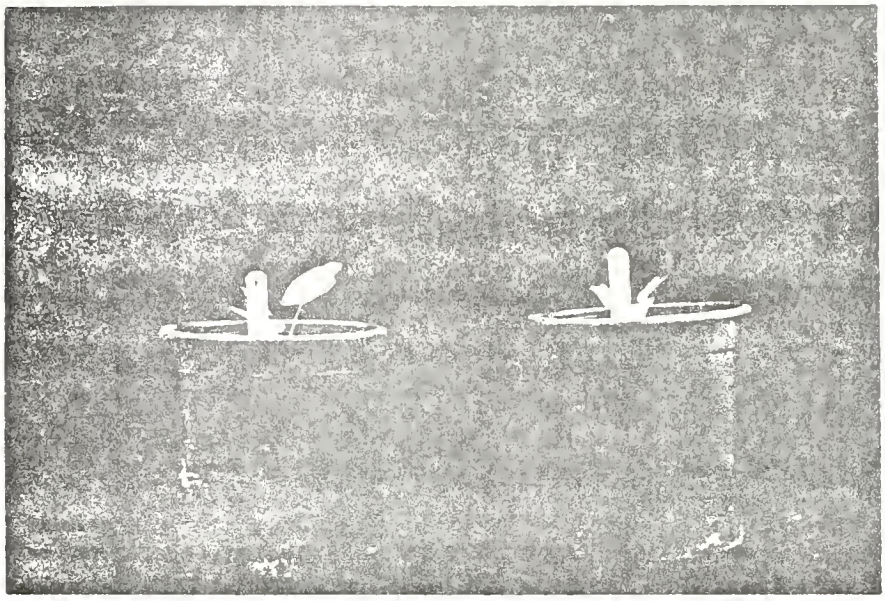
a. Hyphal development and arbuscles (A)



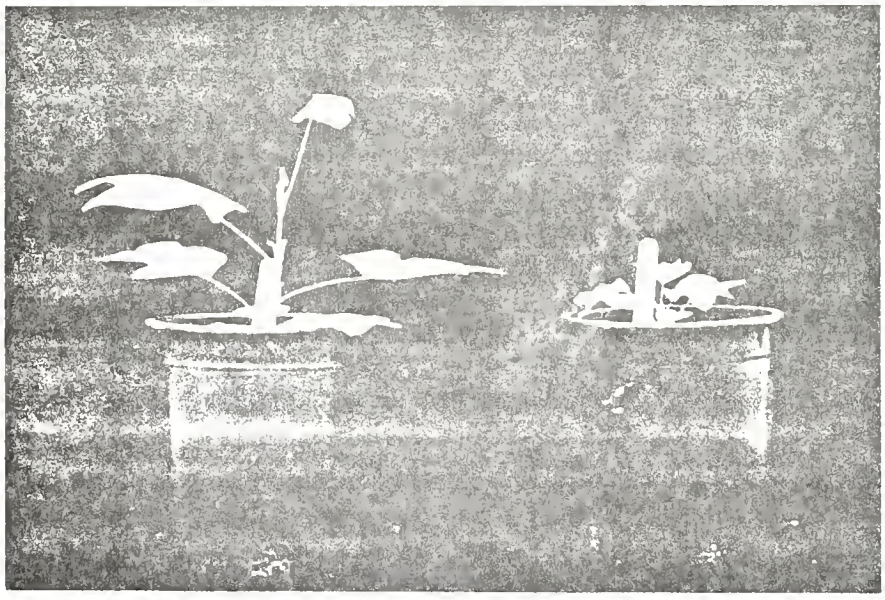
b. Vesicles (V)

Figure 2. Mycorrhizal development by Glomus fasciculatus in roots of Liriodendron tulipifera (72X).

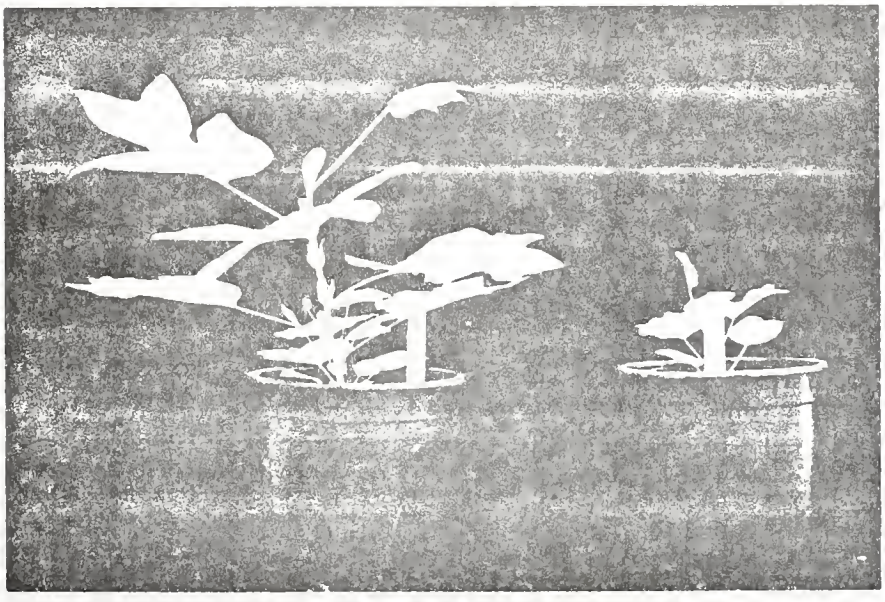
a.



b.



c.



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